

NOV 3 1971

1971 CONFERENCE ON CITRUS CHEMISTRY AND UTILIZATION

OCTOBER 8, 1971

ABSTRACTS OF PAPERS

LANDMARK MOTOR LODGE
Winter Haven, Florida

U. S. Citrus and Subtropical Products Laboratory
600 Avenue S, N.W.
Winter Haven, Florida 33880

SOUTHEASTERN MARKETING AND NUTRITION RESEARCH DIVISION

AGRICULTURAL RESEARCH SERVICE

UNITED STATES DEPARTMENT OF AGRICULTURE

PREFACE

The Conferences on Citrus Chemistry and Utilization are sponsored by the Southeastern Marketing and Nutrition Research Division of USDA's Agricultural Research Service to report research developments in the broad area of processing, marketing, and utilization, and to provide for exchange of information that will benefit the industry and future research.

This report summarizes the statements of the various speakers during the Conference. If further details are desired regarding any subject presented here, they may be obtained by communicating with the author concerned.

C. H. H. NEUFELD, Director
Southeastern Marketing and
Nutrition Research Division

PROGRAM COMMITTEE:

M. K. Veldhuis, Chairman
W. F. Talburt

~~~~~  
Mention of companies or commercial products does not imply recommendation or endorsement by the U. S. Department of Agriculture over others not mentioned.  
~~~~~


PROGRAM

1971 CONFERENCE ON CITRUS CHEMISTRY
AND UTILIZATION

October 8, 1971

THE CHEMICAL EVALUATION OF OFF-FLAVOR DEVELOPMENT IN CITRUS JUICE
Howard L. Dinsmore, Steven Nagy and Harold E. Nordby
U. S. Citrus and Subtropical Products Laboratory
Winter Haven, Florida

DEVELOPMENT OF METHODS FOR PRODUCING CITRUS JUICES FREE OF LIMONIN
BITTERNESS

Vincent P. Maier, Linda C. Brewster and Andrew C. Hsu
Chief, Fruit and Vegetable Chemistry Laboratory
Western Marketing and Nutrition Research Division
Pasadena, California

QUANTITATIVE AND QUALITATIVE ANALYTICAL STUDIES ON SOME CITRUS
ESSENTIAL OILS

Philip E. Shaw, Manuel G. Moshonas and Richard L. Coleman
U. S. Citrus and Subtropical Products Laboratory
Winter Haven, Florida

DIFFERENTIATION OF CLOSELY RELATED CITRUS CULTIVARS THROUGH LIPID
PROFILES

Steven Nagy and Harold E. Nordby
U. S. Citrus and Subtropical Products Laboratory
Winter Haven, Florida

STUDIES ON NONENZYMIC BROWNING IN MODEL SYSTEMS RELATED TO DRIED
CITRUS PRODUCTS

Derek Horton, M. L. Wolf from, and N. Kashimura
Department of Chemistry
The Ohio State University
Columbus, Ohio

CITRUS RIBONUCLEOTIDES AND THEIR POSSIBLE INFLUENCE ON FRESH AND
PROCESSED PRODUCTS

R. H. Biggs
Fruit Crops Department
University of Florida
Gainesville, Florida

YIELD, LIMONIN CONTENT AND FLAVOR RELATIONSHIPS IN ORANGE JUICE

J. H. Tatum, C. J. Wagner, Jr., and R. E. Berry
U. S. Citrus and Subtropical Products Laboratory
Winter Haven, Florida

QUANTITATIVE ANALYSES OF AROMA SOLUTIONS AND CONCENTRATES

E. D. Lund, C. J. Wagner, Jr., and R. E. Berry
U. S. Citrus and Subtropical Products Laboratory
Winter Haven, Florida

CHANGES IN THE TEXAS CITRUS PROCESSING INDUSTRY

Bruce J. Lime
In Charge, U. S. Food Crops Utilization Research Laboratory
Southern Marketing and Nutrition Research Division
Weslaco, Texas

RECOVERY OF VOLATILE ORGANIC MATERIALS FROM ORANGE PEEL

R. E. Berry, C. J. Wagner, Jr., W. L. Bryan and M. K. Veldhuis
U. S. Citrus and Subtropical Products Laboratory
Winter Haven, Florida

SUMMARY OF COMPLETED STUDY OF AIR POLLUTION CREATED BY CITRUS FEED MILLS

John W. Seabury
P. E. Executive Vice President
Fiske-Gay Associates, Inc.
Orlando, Florida

IMPROVED METHOD FOR RECOVERY AND PURIFICATION OF ORANGE PEEL COLOR

C. W. Wilson, III, O. W. Bissett and R. E. Berry
U. S. Citrus and Subtropical Products Laboratory
Winter Haven, Florida

THE CHEMICAL EVALUATION OF OFF-FLAVOR DEVELOPMENT IN STORED CITRUS JUICE

Howard L. Dinsmore, Harold E. Nordby and Steven Nagy

Southeastern Marketing and Nutrition Research Division
Citrus and Subtropical Products Laboratory
Winter Haven, Florida

Off-flavor in citrus products has been considered to arise mainly from temperature-promoted hydration and oxidation reactions on terpene and lipid constituents. While many specific chemical changes have been identified, there has remained a need for an analysis which would correlate closely with off-flavor and moreover would be sensitive enough to detect its onset at a very early stage during storage of bottled juice. We have been investigating two different approaches to finding an off-flavor index: gas-liquid chromatography of juice volatiles and colorimetric analysis. In principle these approaches are opposite. The gas-liquid chromatogram (glc) is virtually a total analysis; ultimately it will show every detail of chemical change during storage. By contrast, a colorimetric method will monitor only one key substance, or class of substances, related in some way to formation of off-flavor; it is not necessary that the index compound be itself an off-odor constituent.

The glc study, which was at an early stage when reported at the 1970 Conference, has been improved and put on a quantitative basis, particularly through better control in the stripping and capture of volatiles. Commercially blended, glass-packed, single-strength orange juice was used in all these studies. Volatiles stripped from 500 ml of freshly opened juice by a stream of nitrogen were captured in a trap of cold solvent, methylene chloride, followed by two empty traps cooled by liquid nitrogen. The combined contents in methylene chloride were reduced on a rotoevaporator to about 0.2 ml for glc analysis. Temperature-programmed chromatograms, run in a uniform manner on 5 microliter samples, have verified the consistency of samples thus prepared, so that changes in glc patterns are real indications of compositional differences in juice samples.

Over 40 peaks have been observed and their individual odors during exit from the chromatograph have been "sniffed." No single peak has had the characteristic off-odor of temperature-abused juice, although certain peaks exiting after limonene lack typically pleasant terpene odors. Of the several peaks (notably those numbered 29, 37 and 39) which became increasingly prominent with long, high-temperature storage, peak 37 showed particular promise of being a sensitive indicator of juice deterioration. The intensity of this peak was measured relative to nearby peak 35 which remained substantially constant, and their consistent ratio of 1.69 for 40°-stored control juice was redefined as "one glc index unit of 37 peak." All relative increases in peak 37 intensity are conveniently expressed in this unit.

It can be seen in Table 1 that all juice samples stored at higher temperatures show an increase in index units roughly proportional to both storage time and temperature. At about 2 index units off-flavor has been observed in most cases.

However, it is hard to assign a precise off-flavor threshold with our limited data. Juice stored two weeks at 85° had 1.88 index units and an unmistakable off-flavor, while juice stored for nine weeks at 60° and having 2.45 index units was just slightly off-flavor, and the nine-week 50° juice with 2.04 index units still tasted good. Probably the need to give more attention to our tasting methods is indicated; but whatever value of glc index may be agreed to show onset of off-flavor, the peak 37 intensity seems to provide an excellent off-flavor indicator. Although compound 37 is more properly related to off-flavor as an index of the level of those compounds which are more directly responsible, its chemical identification would be interesting, and we are now beginning that phase of this study.

In our initial effort to find a colorimetric measure of juice deterioration, we sought to utilize the thiobarbituric acid (TBA) reaction, one of the widely used tests for rancidity in fish, meat and dairy products. Here the key substance is malonaldehyde, an intermediate in the autooxidation of lipid polyunsaturates. This test turned out to be useless with juice. While satisfactory recovery was obtained in distillates from juice spiked with added malonaldehyde, no TBA-reactives were recovered from any natural juice, good or bad; and the necessary acidic conditions for the procedure were borderline to promoting breakdown of the juice by the test, itself.

Other intermediates having simple colorimetric analyses were then considered: oxygenated terpenes, aldehydes, and furfural; and it soon became evident that determination of furfural held the most promise. This compound is totally absent in fresh juice and is considered to develop via breakdown of ascorbic acid. Its presence in juice partly as thiofurfural has been postulated by J. S. Blair at Lake Alfred,¹ who moreover utilized colorimetric determination of furfural as an index of flavor deterioration in canned citrus juice. Since Blair's work was concerned with relatively long storage periods (mostly three months or more) the question still remained of how effectively this determination would indicate onset of off-flavor in bottled juice during the first weeks of storage.

On account of its high vapor pressure, furfural is readily freed and concentrated from very low levels in juice, either by stripping or by distillation. Rapid distillation of the juice in a Scott-Veldhuis oil-determination apparatus has proved to be the most efficient and consistent recovery technique. The first 5% by volume of distillate contains 40% of the total original furfural, a value which we found to be independent of the sample amount and of its furfural level. The concentration of furfural in the distillate is therefore enriched eight-fold relative to the original juice. Working in the range 10^{-3} to 10^{-4} grams of furfural per liter we have moreover obtained exactly the same recoveries from juice spiked with furfural as from simple aqueous solutions.

1

Citrus Experiment Station Mimeo Report 65-4, given at Fifteenth Annual Citrus Processors' Meeting (1964).

2

Citrus Experiment Station Mimeo Report 66-6, given at Sixteenth Annual Citrus Processors' Meeting (1965).

Determination of furfural is possible by many methods, of which direct absorption in the near ultraviolet, color-formation with benzidine, and color formation with aniline are of comparable very high sensitivity. The reaction with aniline gives the least "color blank" or background interference (really none at all) and was clearly superior for recognition of furfural at very low levels. The procedure is rapid and very economical of sample. As little as 2×10^{-4} gram per liter of furfural gives a measurable color. This corresponds to 25 parts-per-billion in the original juice.

Application of furfural determination to juice distillates has given excellent correlation with taste evaluation, especially in detecting the onset of off-flavor, as seen in Table 2. Note that although tasters had difficulty in assigning priority to two of the juices, the chemical results were unambiguous. Table 3 shows how long bottled juice stored at various temperatures stays fresh, on the basis of whether furfural could be detected or not. The subsequent increase in furfural is illustrated in Table 4. We think that this simple analysis provides a valid and sensitive index of bottled juice off-flavor.

Table 1. Gas-Liquid Chromatographic Index of Juice Quality (Peak 37 Units)

Storage °F	Storage Time and Quality		
	2 Weeks	4 Weeks	9 Weeks
40	1.00	1.00	1.00
50		1.19	2.04
60		1.62*	2.45*
70		2.03*	3.20*
85	1.88*	3.09*	3.95*

*

Identifies juices having noticeable off-flavor.

Table 2. Evaluation of Juice After Nine Weeks Storage

Storage °F	PPB Furfural	Taste Rating and Description ¹	
40	None	(Not rated)	
50	None	1, 2	Good taste
60	30	1, 2	Good taste
70	50	3	Mild off-flavor
85	200	4	Considerable off-flavor

¹ Numbers increase with off-flavor; tasters varied in their ability to distinguish the 50° from the 60° juice.

Table 3. Juice Stability as Evaluated by First Discernible Furfural¹

Storage °F	Onset of Noticeable Furfural
40	Well over 1 1/2 years
50	About one year
60	Nine weeks
70	Six weeks
85	Under two weeks

¹ About 30 parts-per-billion

Table 4. Furfural Increase in Juice With Storage Time

Weeks	At 70°F	At 85°F
2	None	40 ppb
4	Maybe trace	80
7	30	160
9	50	200

DEVELOPMENT OF METHODS FOR PRODUCING CITRUS JUICES

FREE OF LIMONIN BITTERNESS

Vincent P. Maier, Linda C. Brewster and Andrew C. Hsu

Western Marketing and Nutrition Research Division
Fruit and Vegetable Chemistry Laboratory
Pasadena, California 91106

Despite the overall high quality of the citrus crop, portions of the crop tend to yield juice whose quality suffers because of limonin caused bitterness. While limonin bitterness has historically been associated with navel orange juice, it is now recognized that the juice of other varieties of oranges as well as that from lemons, grapefruit and tangerines is on occasion subject to limonin bitterness. This type of bitterness is unique in that it generally develops after the juice is extracted from the fruit. In earlier work we have shown that the change in taste is caused by the conversion of a nonbitter substance (limonoate A-ring lactone) to a bitter substance (limonin) after the juice is extracted from the fruit (Maier and Beverly, 1968; Maier and Margileth, 1969). The nonbitter substance occurs naturally in the fruit tissues where it is stable and remains nonbitter. However, when the fruit tissues are broken during juice extraction this normally nonbitter substance is attacked by the juice acids and is converted into the intensely bitter substance limonin. Juice containing as little as six parts per million of limonin is bitter.

Up to the present time no practical method has been found to remove limonin from juice. However, research on the chemistry and biochemistry of limonin over the past several years has opened two promising new approaches to this problem, namely, metabolic debittering (Maier and Brewster, 1971) and enzymatic debittering (Bennett and Hasegawa, et al. 1971). Both approaches are under investigation at our laboratory in Pasadena. In this report primarily the work on metabolic debittering will be discussed.

Studies on the biochemistry of limonin led to the observation of a metabolic system in the intact orange that functions, while the fruit is still on the tree, to prevent bitterness from developing later in the juice. However, this system has its effect late in the harvest season after most of the navel orange crop has been harvested. It was found that this metabolic system acts to prevent bitterness by destroying the nonbitter precursor substance. Based on these observations it was reasoned that the juice bitterness problem could be solved if a way could be found to activate this natural metabolic system at an earlier stage of fruit maturity. In biochemical language an agent was needed to "turn-on" the system.

A search for such agents is underway. One agent which has been found to be effective is 2-chloroethylphosphonic acid (CEPA) (Maier and Brewster, 1970

and 1971). We have found that within five days after the fruit has been dipped in a dilute water solution of CEPA the limonin content of the extracted juice is 20 to 40 percent lower than that of the juice from fruit before the treatment. Taste tests confirm that the treatment results in juice of substantially reduced bitterness while at the same time the juice retains the desirable flavor characteristics of fresh orange juice.

While CEPA appears to have considerable promise it is not yet certain whether it will be the agent which is eventually adopted for use by the citrus industry. Before it can be used commercially CEPA must be thoroughly tested and proven safe for use on foods, as required by the FDA. Such tests are currently being conducted by the manufacturer.

Consequently, we are continuing our search for other agents, based on what we have learned about the mechanism for "turning-on" the debittering metabolic system. We now know that CEPA acts through a specific ethylene mechanism which appears unrelated to the general effects this substance has on plants. (Maier, Brewster and Hsu, 1971). Prolonged, accelerated respiration, a common ethylene effect, is not required for the debittering metabolic system to function. In fact exposure of the fruit to low concentrations of ethylene for as brief a period as three hours is sufficient to trigger the metabolic debittering system. Thus, navel oranges treated with 20 ppm ethylene for three hours followed by a five day holding period with no ethylene produced juice having a 32% lower limonin content than the juice from the fruit before treatment. In the same experiment CEPA caused a similar reduction in juice limonin levels. Exposure of the fruit to ethylene for the entire five day period gave lower quality juice while at the same time producing no greater effect on limonin levels than the three hour treatment.

The triggering of metabolic debittering by brief exposure of the fruit to ethylene is a new ethylene effect that has considerable commercial potential. Further work is in progress to determine the optimum conditions of treatment, however, there appears to be no reason why commercial application of the approach (treatment of the fruit for several hours with 20 ppm ethylene, removal from gassing room followed by a holding period of about five days before juicing) to producing citrus juices of reduced limonin bitterness could not be begun now on a trial basis.

Other research on the limonin bitterness problem is being conducted by Hasegawa and Bennett at our laboratory. The objective of this work is to produce a specific enzyme which can be added to bitter juice to destroy limonin. It is envisioned that success in both approaches will lead to an eventual two-pronged system whereby accelerated metabolism greatly reduces the amount of limonin entering the juice and the debittering enzyme destroys any which does enter. Thus far, one limonin degrading enzyme from microbial sources has been isolated and the isolation of others is in progress. Such enzymes also hold promise for use in the development in a rapid limonin assay procedure for citrus juices.

QUANTITATIVE AND QUALITATIVE ANALYTICAL STUDIES ON SOME CITRUS ESSENTIAL OILS

Philip E. Shaw, Manuel G. Moshonas and Richard L. Coleman

Southeastern Marketing and Nutrition Research Division
Citrus and Subtropical Products Laboratory
Winter Haven, Florida

Quantitative analytical studies of citrus essential oils have generally relied on some preliminary separation before quantitative gas chromatographic (glc) analysis of the desired fraction is performed (Lifshitz et al. 1970 and references therein). However, Bernhard (1960) quantitatively analyzed cold-pressed lemon oil by glc without a preliminary separation step, but neither the amount of nonvolatile material present, nor glc response factors were determined in that study.

We became interested in analyzing citrus oils quantitatively during studies comparing the composition of Valencia orange essence and aroma oils and later, grapefruit essence and aroma oils. Essence oil is the oil phase separated during the preparation of commercial aqueous essence and aroma oil is the oil phase separated after distillation of the aqueous discharge from peel oil centrifuges (Veldhuis et al., 1970). Comparison of Valencia orange essence and aroma oils has already been reported (Coleman and Shaw, 1971).

Quantitative studies on grapefruit essence and aroma oils were carried out by glc separation to give the oil components listed in Table 1. Quantities of each component present were determined by relating the area of each peak present to total area under the curve. No response factors were determined in this study since the primary purpose was to compare essence oil and aroma oil composition. For more accurate results, glc response factors would have to be determined for each component by injecting a known amount of that component into the same column under conditions used for the oil analyses. Distilled essence and aroma oils can be injected directly into a glc without prior separation because the waxes and other high-boiling components found in cold-pressed oils are not present in these distilled oils. From the qualitative and quantitative composition as given in Table 1, one can see that there are no major differences in composition between these grapefruit essence oil and aroma oil samples.

Cold-pressed citrus oils are not as amenable to quantitative analysis by glc as are the distilled essence and aroma oils, because of the waxes, carotenoids and other high-boiling components present in the cold-pressed oils. For meaningful quantitative glc results using cold-pressed citrus oils, the quantity of nonvolatile material present must be determined. Furthermore, some means of removing this nonvolatile material from the glc column must be available if a column is to be used for repeated analyses of oil samples.

Table 2 lists the quantitative results of Persian lime oil analysis by a procedure that permits both the percentage of nonvolatile material to be determined and the glc column to be regenerated for subsequent use. Molecular still distillation of Persian lime oil samples at reduced pressure has repeatedly shown nonvolatile material to be 7.5% of the total oil. Removal of nonvolatile material from the glc column was accomplished by injection of one or more small quantities of the silanizing agent Silyl 8 (Pierce Chemical Co., Rockford, Ill.). This silanizing treatment not only stripped nonvolatiles from the column in 5-10 minutes, but regenerated the column so that the difficult separation of β -caryophyllene and bisabolene was possible on the following run. Without silanizing between each run, column resolution suffered.

The same technique for quantitative analysis can be applied to other cold-pressed citrus oils as well. Preliminary results with cold-pressed Valencia orange oil have shown that a longer column than the 6-ft. column used in the lime oil work is needed for adequate separation of the main components from this oil.

Meyer lemon peel oil was qualitatively analyzed and some 31 components were identified as listed in Table 3. The first alcohol listed, thymol, comprised about 6% of the total oil. Its flavor threshold in water is about 1.7 ppm, and thus this component was present in the oil in an amount far exceeding its threshold. The medicinal-type odor (Lavoris mouthwash) was judged objectionable to the taste panel if present in citrus juices. Other noteworthy differences between Meyer and other lemon oils were its high limonene content and its very low citral (neral + geranial) content.

Meyer lemon peel oil was reported by Wenzel et al. (1958) to cause off-flavor in lemon juice if the juice is not thoroughly deoiled before being used. The present study indicates that if Meyer lemon oil is to be a commercially valuable product, use should be made of its thymol-like odor and flavor rather than its potential to produce a lemon-like flavor.

Literature Cited

- Bernhard, R. A. 1960. Analysis and Composition of Oil of Lemon by Gas-Liquid Chromatography. *J. Chromatog.* 3: 471-476.
- Coleman, R. L. and Shaw, P. E. 1971. Analysis of Valencia Orange Essence and Aroma Oils. *J. Agr. Food Chem.* 19: 520-523.
- Lifshitz, A., Stanleh, W. L. and Stepak, Y. 1970. Comparison of Valencia Essential Oil from California, Florida and Israel. *J. Food Sci.* 35: 547-548.
- Veldhuis, M. K., Berry, R. E. and Wagner, C. J., Jr. 1970. Preparation and Use of Concentrated Orange Aroma Solutions. 1970 Conference on Citrus Chemistry and Utilization, Winter Haven, Fla., Oct. 9.
- Wenzel, F. W., Olsen, R. W., Barron, R. W., Huggart, R. L., Patrick, R. and Hill, E. C. 1958. Use of Florida Lemons in Frozen Concentrate for Lemonade. *Proc. Fla. State Hort. Soc.* 129-132.

Table 1. Quantitative Estimation of Whole Grapefruit Essence and Aroma Oils

Compound	R.T. (min) ^a	% Area Under Curve	
		Essence Oil ^b	Aroma Oil
Heptane	3.0	-	< .01
Acetone	4.2	-	< .01
Ethanol	8.4	0.03	-
α -Pinene	13.8	0.05	0.15
Myrcene	20.4	-	-
D-Limonene ^a	26.4	99.18	98.50
Octanal	27.6	0.09	0.37
Nonanal	33.0	0.03	0.02
Decanal	38.2	0.05	0.23
Linalool	39.4	0.08	0.18
β -Caryophyllene	45.0	0.09	0.06
α -Terpineol	47.8	0.07	0.07
Geranial	49.8	0.10	0.07
Carvone	51.0	-	< .01
Valencene	51.0	0.10	0.06
Δ -Cadinene	52.2	0.02	0.05
Nootkatone	103.0	0.04	0.03

^a A 0.25 in. by 20 ft. column packed with 20% Carbowax 20M on 60-80 mesh Gas Chrom P.

^b Redd Laboratories, Inc., Lakeland, Fla.

Table 2. Quantitative Analytical Data for Persian Lime Oil

Compound	Glc Area %		Synthetic mixt		Corrected Wt % of	
	Lime oil	Synthetic	Mg added	Calc wt%	Volatiles	lime Oil
α -Pinene	2.75	2.99	13.79	2.87	2.59	2.40
β -Pinene	10.95	9.62	55.24	11.31	12.63	11.68
D-Limonene	56.38	64.28	282.12	58.85	50.65	46.85
γ -Terpinene	14.30	12.93	76.23	15.90	17.25	15.96
Terpinolene	1.12	1.94		1.18	0.66	0.61
Neral	2.32	1.14	8.06	1.68	3.35	3.10
Geranial	4.54	4.10	23.79	4.96	5.38	4.98
Neral Acetate +	2.72	2.27	13.19	2.75	3.23	2.99
Geranyl Acetate						
β -Caryophyllene +	1.11	0.81	5.76	1.19	1.59	1.47
Bergamotene						
β -Bisabolene	2.12	1.81	10.93	2.28	2.62	2.42

Table 3. Compounds Identified in Meyer Lemon Peel Oil

ALCOHOLS

Thymol
 Linalool
 α -Terpineol
 Citronellol
 Nerol
 1-p-Menthene-9-ol
 1,8-p-Menthadiene-9-ol
 Terpinen-4-ol
 p-Cymene-8-ol

ALDEHYDES

Citronellal
 Neral
 Geranial

ESTERS

Citronellyl Acetate
 Neryl Acetate
 Geranyl Acetate
 1,8-p-Menthadien-9-yl-acetate

HYDROCARBONS

Limonene
 γ -Terpinene
 p-Cymene
 Terpinolene
 α -Pinene
 β -Pinene
 Myrcene
 α -Cubebene
 α -Copaene
 α -trans-Bergamotene
 α -Selinene
 Farnesene
 β -Elemene
 β -Bisabolene

MISCELLANEOUS

o-Phenyl phenol

Table 1. Long-Chain Hydrocarbon Profiles of Citrus

Carbon No.	Valencia			Duncan			King		
	N	I	AI	N	I	AI	N	I	AI
20	0.19	-	t	0.26	t	t	0.37	t	.01
21	0.71	t	t	0.36	0.02	t	0.48	0.03	t
22	2.60	0.09	t	1.69	0.10	0.05	1.58	0.10	0.03
23	17.37	10.08	0.15	12.48	10.15	0.23	12.17	6.06	t
24	5.92	3.39	7.19	6.16	2.70	6.71	5.53	1.75	6.85
25	14.93	12.76	1.37	13.53	14.67	2.05	17.83	11.31	2.21
26	1.64	1.46	5.56	2.60	1.53	5.96	2.43	0.97	8.72
27	2.77	3.19	0.40	4.58	4.37	0.59	3.89	4.76	1.02
28	0.50	0.15	1.50	1.00	0.40	1.75	0.58	0.35	3.85
29	1.41	0.68	0.04	1.64	1.14	0.24	1.05	1.11	0.24
30	0.50	0.02	0.40	0.54	0.21	0.57	0.48	0.20	1.10
31	1.05	0.24	0.11	0.94	0.33	0.10	0.88	0.37	0.15
32	0.48	0.01	0.19	0.20	0.02	0.12	0.22	0.07	0.28
33	0.39	0.12	t	0.35	0.12	t	0.19	0.08	0.04
34	0.23	t	t	0.08	t	.03	0.09	0.03	0.01
35	0.17	t	t	0.11	t	t	0.10	t	t

Table 2. Major Fatty Acids of Triglycerides (%)

Acid	Hamlin	Valencia	Temple
Palmitic	10.51	7.67	8.21
Palmitoleic	7.04	7.79	8.53
Oleic	33.22	29.90	32.24
Linoleic	28.93	35.54	13.02
Linolenic	16.44	16.16	34.19

Table 3. Major Fatty Acids of Monogalactosyl Diglyceride (%)

Acid	Hamlin	Valencia	Temple
Palmitic	5.26	6.69	6.36
Palmitoleic	6.49	6.14	7.54
Oleic	27.11	27.06	31.02
Linoleic	19.07	26.44	6.86
Linolenic	40.02	31.21	44.61

STUDIES ON NONENZYMIC BROWNING IN MODEL SYSTEMS RELATED TO DRIED CITRUS PRODUCTS

Derek Horton, M. L. Wolfrom, and N. Kashimura

Department of Chemistry
The Ohio State University
Columbus, Ohio

This report describes a study on the development of new analytical tools for monitoring the chemical transformations that take place in the nonenzymic browning reaction between sugars and amino acids, with particular emphasis on the chemical processes involved in the deterioration of dried citrus juices upon storage. The results obtained with model systems of sugars, amino acids, and other constituents are compared with those found when the techniques are applied to samples of dried orange-juice powder produced by the foam-mat process. The key intermediate in the browning reaction, 3-deoxy-D-erythro-hexosulose, and the chemical reactions leading to its formation have been examined in some detail.

With the eventual objective of setting up a fully automated system, based on gas-liquid chromatography--mass spectrometry (g.l.c.-m.s.), for separation, quantitation, and structural identification of components present in the complex mixtures formed when sugars and amino acids interact in food products to cause product deterioration through the sequence of reactions known as nonenzymic browning (Maillard reaction),¹ a study has been made (a) on the volatilization and separation by g.l.c. of the intermediates in the browning reaction, and (b) examination by m.s. of the volatile derivatives.

After evaluation of a range of possible reagents for converting sugars and their transformation products in the browning reaction into volatile derivatives suitable for separation by g.l.c., it was determined that trimethylsilylation by use of 5:5:1 (v/v) mixture of N, O-bis(trimethylsilyl)acetamide, N-trimethylsilylimidazole, and chlorotrimethylsilane was the most effective system.² With this reagent it was found possible to obtain reproducible g.l.c. peaks, suitable for quantitative work, with D-glucose, D-fructose, and various glycosylamines, together with their Amadori transformation products formed in the reaction of D-glucose with p-toluidine, 4-aminobutyric acid, and various α -amino acids. Reproducible peaks were also observed for the amines and amino acids present in the mixtures.

The trimethylsilylated products from the foregoing experiments were examined by infrared and mass spectrometry. The mass spectra were interpreted to confirm the structures assigned, and were also used as "fingerprints" to identify the products separated from the mixtures by preparative g.l.c.

By use of the trimethylsilylation technique so developed, analyses were made as a function of time and temperature of reaction mixtures set up as model systems for the nonenzymic browning reaction. The examples studied initially included simple systems of sugars and amino acids or amines, with appropriate variations of the nature and concentration of reactants. The technique permits monitoring of the loss of D-glucose from the system, formation of the browning intermediates, and their further transformation into other products. Detailed studies were then made in a system selected as a model for the dried citrus juice, containing D-glucose, citric acid, potassium citrate, sodium benzoate, L-ascorbic acid, and an amino acid (either glycine, 4-aminobutyric acid, L-arginine, or L-lysine). The procedure was applied to the samples of orange-juice powder that had been stored under various conditions of time and temperature, and direct correlations with the model systems were made.

The techniques developed provide a simple and rapid method for the objective detection and quantitation of the extent of deterioration caused by nonenzymic browning in citrus and other food products. Furthermore, the technique furnishes a useful tool for detailed chemical studies on the transformations involved in the browning reaction. For this purpose, attention has been focused on the mode of formation and subsequent decomposition of the key intermediate in the browning reaction, namely 3-deoxy-D-erythro-hexosulose³, a product formed by the sequence D-glucose + amino acid \rightarrow N-substituted D-glucosylamine \rightarrow 1-N-(amino acid)-1-deoxy-D-fructose \rightarrow 3-deoxy-D-erythro-hexosulose. A convenient new preparative route⁴ has made this key intermediate readily available for detailed study: other routes to this compound were examined and will be mentioned briefly. In a study (with D. Tam) on the formation of 3-deoxy-D-erythro-hexosulose by use of deuterium-labeled reagents it was found that one deuterium atom is introduced at C-3; the significance of this result on the mechanism of the reaction leading to the hexosulose will be discussed.

LITERATURE CITED

1. L. C. Maillard, Compt. Rend., 154, 66 (1912); Ann. Chim., [9] 5, 258 (1916); T. M. Reynolds, Advan. Food Res., 12, 1 (1963); 14, 168 (1965).
2. M. L. Wolfrom and N. Kashimura, Carbohydr. Res., 11, 151 (1969).
3. E. F. L. J. Anet, Aust. J. Chem., 13, 396 (1960); 18, 240 (1965).
4. H. El Khadem, D. Horton, M. H. Meshreki, and M. A. Nashed, Carbohydr. Res., 17, 183 (1971).

CITRUS RIBONUCLEOTIDES AND THEIR POSSIBLE INFLUENCE ON FRESH AND PROCESSED PRODUCTS

R. H. Biggs

Fruit Crops Department
University of Florida
Gainesville, Florida

Isolation and chemical analyses have been made of nucleosides, nucleotides and nucleic acids of several cultivars of citrus, including oranges, tangelos and grapefruits. Particular attention was given to patterns of changes within nucleotides and nucleic acids as influenced by growth, stage of maturity, handling of fresh fruits and extracted products, storage conditions, and exposure to ethylene.

For separation and identification of nucleosides and nucleotides from citrus fruits, methods used with other tissues had to be slightly modified. Briefly outlined are the procedures that were used as a standard for this investigation: extraction with perchloric acid, subjection to filtration through insoluble polyvinyl-pyrrolidone, adsorption of nucleosides and nucleotides on activated charcoal, desorption with ammoniacal, 50 percent ethanol and ion exchange chromatography for initial separation, with subsequent separation by thin-layer and gas chromatography. Comparisons will be presented of nucleoside and nucleotide patterns obtained with various isolation procedures. Also, patterns obtained from fruits of various citrus cultivars after the designated treatments.

For extraction, separation and tentative identification of the nucleic acids, the following briefly outlined procedure was used as the standard: grind frozen tissue (-70 C) in a phenol: aqueous media, centrifuge, retain aqueous phase after suitably washing, precipitate RNA with cold ethanol, centrifuge, redissolve precipitate, reprecipitate, centrifuge and wash pellet. The RNA pellet was then redissolved and separated further by gel electrophoresis. Tentative identification was by ultra-violet spectrophotometric analyses of the components on the electrophoretogram and the rate of incorporation of ^{32}P from ortho-phosphate by the juice vesicles. Using this procedure, subunits of ribosomal particles (23S and 18S) and soluble RNA's of fruits have been analyzed. Patterns of changes of polymeric nucleotides will be discussed as related to development, stage of maturity and handling practices.

It would seem that both acid soluble components and polymeric nucleotides are in a dynamic state in fruits, with synthesis and interconversions being influenced by stage of growth and various environmental factors. Concentrations in fruit tissues and patterns of change of nucleotides and nucleic acids indicate they play a major role in quality of fresh fruits and processed products. In addition to the role of these components in cellular reactions, the concentration present in fruits of certain of the nucleotides could have a direct modifying effect on the taste of products through giving "body" feel and indirect through modifying other flavor components.

YIELD, LIMONIN CONTENT AND FLAVOR RELATIONSHIPS IN ORANGE JUICE

J. H. Tatum, C. J. Wagner, Jr., and R. E. Berry

Southeastern Marketing and Nutrition Research Division
Citrus and Subtropical Products Laboratory
Winter Haven, Florida

Juice samples were prepared from different orange cultivars (Hamlin, Pineapple and Valencia) using relatively low and high extraction pressures. Thirty boxes of each type fruit were randomized and 15 boxes each were used for a low yield and a high yield juice run. Juice yield was controlled through changes made at the extractor. All juices were finished on a cylindrical screw finisher set with 12 lbs. (Hamlin) to 15 lbs. (Pineapple and Valencia) air pressure using a screen with 0.033 in. hole diameter. All juices were concentrated in a pilot plant falling film evaporator at 3 to 10 mm Hg. Yields (%) were calculated on the basis of unfinished juice/pound of fruit.

Juices were evaluated on a basis of limonin content (Maier, 1970), recoverable oil, and flavor. Yields, limonin levels and oil levels are shown in Table 1.

Table 1

	Yield %		Limonin ppm		% Oil on	
	Low	High	Single	Strength	Reconstituted	Basis
			Low	High	Low	High
Hamlin	54.1	66.8	1.4	5	0.005	0.01
Pineapple	58.8	74.2	1.0	14	0.04	0.07
Early Valencia	55.5	69.4	0.03	5.5	0.01	0.035
Late Valencia	57.6	79.8	0.87	7.3	0.027	0.04

All taste tests were triangulation type, comparing low yield to high yield samples for detectable differences. The oil levels of the low yield juices were adjusted to the high levels and taste tests carried out. There were significant differences between low and high yield samples in all four runs. The low yield juices were preferred by our taste panel. As shown in Table 1 the oil level in the Pineapple juice was too high for objective taste testing. The Hamlin, early Valencia and late Valencia low yield juices were adjusted to the same oil levels and limonin contents as the high yield juices. In all three cases taste tests showed a significant difference still existed, and the low yield juices were still preferred by our panel.

A sample of the early Valencia low yield juice was divided and half was used as a control. To the other half limonin was added to 5.5 ppm and taste tests showed a significant difference, with the control being preferred. This procedure was repeated on several commercial concentrates and there was a significant difference in each case with the control being preferred. These taste test results indicate that limonin does affect the flavor of orange juice when its level is high enough but it was not the entire problem with the high yield juices. Most tasters referred to the greenish, astringent, woody or immature flavors in high yield juice whereas limonin usually causes a pronounced bitterness. Studies are presently underway to determine if the components that cause these undesirable flavors can be identified. Most of these results are based on a single set of experiments and further studies are underway.

QUANTITATIVE ANALYSES OF AROMA SOLUTIONS AND CONCENTRATES

E. D. Lund, C. J. Wagner, Jr., and R. E. Berry

Southeastern Marketing and Nutrition Research Division
Citrus and Subtropical Products Laboratory
Winter Haven, Florida

A study was made of the quantitative composition of aroma solutions and the relationship of type and maturity of fruit, and storage conditions of peel. Quantitative analyses were carried out on aqueous aroma solutions produced from Hamlin, Pineapple, and Valencia orange peels at various stages of maturity. Aroma solutions from frozen and stored Valencia peel were also analyzed for comparison.

The analytical technique used was the solvent extraction-GLC method of Wolford and coworkers for the less-water soluble organics, combined with a separate GLC analysis for ethanol, methanol, and acetaldehyde on a Porapak column as previously described at last year's conference.

Linalool, Limonene, α -terpineol and cis-3-hexene-1-ol were the major components of all the aroma solutions from peel extracts as with the previously reported aroma solutions prepared from Valencia peel-mill desludger effluent. Considerably larger quantities of n-hexanal, 1-hexanol, t-2-hexene-1-ol, t-2-hexenal, 1-octanol, n-octanal, neral, geranial, 1-pentene-3-ol and ethyl vinyl ketone were found in some of these aroma solutions than had been found previously in those from Valencia effluent.

Most of the aroma solutions contained about 20 identifiable components. Table 1 lists all compounds isolated from the various aroma solutions in this study.

The aroma solutions prepared from frozen, stored, Valencia peel contained smaller quantities of the aldehydes n-hexanal, t-2-hexenal and n-octanal and larger quantities of the corresponding alcohols c-3-hexene-1-ol, 1-hexanol, and 1-octanol relative to the usual Valencia peel-derived product. The product from frozen, stored peel differed in several other respects. A large peak tentatively identified as ethyl vinyl ketone was observed in the GLC trace. The corresponding reduced compound, 1-pentene-3-ol was present in relatively small concentration. Ethanol and methanol concentrations were unusually high, while the acetaldehyde concentration was in the normal range. These data suggest that the aldehydes were being reduced to alcohols (probably enzymatically) under the storage conditions used.

The most striking difference among samples from different stages of maturity was the large variation in the relative amounts of the 6-carbon compounds in aroma solutions from Hamlin orange. The immature peel aroma solution (11-4) contained large amounts of t-2-hexene-1-ol, t-2-hexenal and 1-hexanol and relatively small quantities of c-3-hexene-1-ol and n-hexanal. In the more mature Hamlin peel aroma (12-18), c-3-hexene-1-ol was the major 6-carbon

compound which is similar to the typical composition of Pineapple and Valencia peel aroma solutions. In the very mature Hamlin (2-17) aroma solutions there were unusually large quantities of 6-carbon compounds, and c-3-hexene-1-ol, t-2-hexenal and 1-hexanol predominated. These were accompanied by a moderate amount of n-hexanal and a very small quantity of t-2-hexene-1-ol.

The variations observed in the concentrations of these 6-carbon compounds and in the aldehyde-alcohol ratio probably have a pronounced effect on the flavor quality. Probably the aldehydes have an important role in desirable flavor quality and the 6-carbon compounds contribute to greenish, immature aspects of the flavor.

Table 1. Peel Aroma Constituents

Compounds	Concentration Range Wt. % X 10 ³
Methanol	12-344
Acetaldehyde	3-74
Ethanol	24-1200
Linalool	0.9-3.5
α -Terpineol	0.4-1.8
Cis-3-hexene-1-ol	0.16-1.4
Limonene	0.08-1.1
n-Octanal	0.06-0.3
l-Hexanol	0.06-0.8
l-Octanol	0.07-1.3
t-2-Hexenal	0.02-0.8
Terpinene-4-ol	0.04-0.18
n-Hexanal	0.03-0.2
Nerol	0.05-0.24
Geraniol	0.02-0.4
Neral	0.10-0.27
Geranial	0.10-0.31
c-Carveol	0.02-0.03
t-Carveol	0.01-0.12
c-Linalool oxide	0.02-0.04
t-Linalool oxide	0.0006-0.025
l-Pentene-3-ol	0.05-0.52
n-Amyl Alcohol	0.03-0.07
n-Decanal	0.01-0.03
t-2-Hexene-1-ol	0.02-0.75
Terpinyl Formate	0.08
Ethyl-3-Hydroxyhexanoate	0.11-0.24
c-2,8-p-Menthadiene-1-ol	0.02-0.07
t-2,8-p-Menthadiene-1-ol	0.05-0.12
l,8-p-Menthadiene-9-ol	0.05-0.18
Δ -8(10)-p-Menthene-1,2-diol	0.12
Citronellol	0.02-0.13
Perilla Aldehyde	0.02-0.04

CHANGES IN THE TEXAS CITRUS PROCESSING INDUSTRY

Bruce J. Lime

Southern Marketing and Nutrition Research Division
Food Crops Utilization Research Laboratory
Weslaco, Texas

Texas citrus production reached a high of approximately 28 million boxes of fruit in the middle and late 1940's. Due to a series of weather reversals the production has fluctuated since 1950 from almost zero to 16 million boxes in 1970-71. Although a severe freeze occurred in 1962, the 1970-1971 production figure represents an approximate 50% increase in production over the 1960-61 season. During these periods of fluctuating production single strength juice accounted for almost 100% of the processed citrus products. Pryor to the mid 1950's processing utilized from 20-25% of the Texas citrus crop. Today processing utilizes approximately 50% of the crop. The production of orange concentrate has been the major contributing factor to the increase in processing.

Hamlin, Marrs (earlies), and Valencia (late) are the three varieties of oranges grown in Texas. All three varieties produce juices having sufficient color for Grade A single strength juice during the period when the Brix to Acid ratio meets Grade A standards, but only juice from Valencia oranges will meet the Grade A color requirement for Frozen Concentrated Orange Juice. Valencia oranges usually mature from 16 to 18 weeks later in the season than the early orange varieties, however, there is a 4 to 6 week period of overlap when low ratio Valencia juice may be used to blend with high ratio early orange juice to produce an acceptable Grade A single strength orange product. The increased evaporative capacity recently installed by the Texas Citrus Industry will make it possible to upgrade color and adjust the B to A ratio of concentrate produced from Texas early oranges by blending with Texas Valencia orange juice.

RECOVERY OF VOLATILE ORGANIC MATERIALS FROM ORANGE PEEL

R. E. Berry, C. J. Wagner, Jr., W. L. Bryan and M. K. Veldhuis

Southeastern Marketing and Nutrition Research Division

Citrus and Subtropical Products Laboratory

Winter Haven, Florida

Volatile organic materials (aqueous aroma solutions and distilled aroma oils) were recovered from three different types of peel feedstocks by stripping and distillation methods which were described at last year's Citrus Conference. Feedstocks were introduced into a stripper where they were contacted with steam, the vapors were then passed through a side arm into a glass distillation column packed with stainless steel Raschig rings. Below the point at which vapor was introduced into the column, was a short stripping section where the liquid condensation was stripped counter-current with steam from a reboiler at the bottom. Stripped vapors then passed through an upper fractionating and rectification section, through a warm, partial condenser (to provide reflux), to a cold condenser and a receiver. The receiver was arranged so that the water and oil phases of the final product could be collected separately.

Three types of feedstocks were studied in this system. These were: 1) commercial centrifuge effluent obtained from the desludging centrifuges at a commercial peel oil mill, 2) pilot plant prepared peel oil emulsions obtained by pressing Valencia peel from a Brown extractor through a screw press with a water rinse and passing the resultant slurry through a finisher to provide a final emulsion containing about 95-6% water, 3-4% solids, and 1% oil, (Studies indicated this pilot plant procedure removed only 18% of the oil from the peel), 3) water extract of ground peel obtained by grinding Valencia peel from AMC extractors with a Hobart food grinder (0.177-in diameter openings) followed by a hammer-mill (0.125-in diameter openings). The ground peel was slurried with chilled water for 30-min and then sufficient water added to make a fluid mixture which was finished as in 2. This provided an oil emulsion containing about the same water-solids-oil composition as 2 but resulted in removing about 73% of the oil from the original peel, primarily through contacting more finely ground peel with larger quantities of water for longer times. For comparison of these feedstocks about 10 to 15% of the feed stream was vaporized and fed to the fractionating column for concentrating the products. Aroma solutions were analyzed for total recoverable organics and flavor potential and feedstocks were compared by aroma solution analyses and oil recovery.

Several methods of contacting steam with the feed liquid were compared for efficiency of stripping. These included: 1) a feed liquid spray nozzle, 2) turbulent-flow long tube contactors of 1-in and 2-in diameter, 3) a steam nozzle with 1/8-in opening (stainless steel), and 4) a copper tubing steam coil 30-in long with 155 holes varying from 1/64-in to 1/32-in diameter. In the comparison of 1 and 2 up to 90% oil recovery could be obtained with the

spray nozzle while only 80 to 85% recovery could be achieved with the long tube contactors, however, organoleptic evaluation of the aroma solutions and oils appeared to indicate higher quality from the long-tube contactors. When the steam nozzle (3) was used, oil recoveries of about 69, 85 and 90% were obtained for respective vaporizations of about 3, 11, and 17%. When the steam coil (4) was used, recoveries of about 79, 82, and 84% were obtained for respective vaporizations of about 8, 13 and 16%. These systems are still under evaluation but preliminary indications are that the steam nozzle is more efficient than the steam coil and that quality of products may be enhanced with the long tube contactors (possibly due to shorter residence times).

Several runs were carried out to demonstrate the operation of the column at total reflux, recycling the aqueous phase product to the top reflux section of the column in order to achieve maximum enrichment of the organics in the oil phase product. Several runs were made with this arrangement, but methanol, ethanol and alcohol-free COD measurements, on the material being recycled to the enrichment section, indicated equilibrium had not been reached even after six hours of operation. As expected, these indicators increased with operation time but it was not expected that they should still have not leveled off after seven hours. Further tests are being planned in this study.

Another experiment was carried out to determine whether aroma solutions and aroma oils could be obtained directly from peel by application of heat and vacuum. Valencia peel was ground on a Hobart grinder as described above yielding a material with 28% solids, 70% moisture and about 2% oil. Several pounds of this material were placed in a pilot plant rotating vacuum evaporator operated with 122°F heating bath, 15 rpm, 48°F condenser under a pressure of less than 0.1 mm Hg. Product was collected in dry ice and liquid nitrogen traps as well as a chilled water receiver. Seven hours operation resulted in 32% weight loss and in a reduction of oil content from 6.9% oil (solids basis) in the feed to 2.7% oil (solids basis) in the waste. The recovered volatiles were distributed 85% in the cold water receiver, slightly less than 15% in the ice trap and less than 0.5% in the liquid nitrogen trap. About 12% of total aqueous distillate and 77% of the total oil phase was collected in the dry ice trap. Analyses indicated a distribution of methanol, ethanol and alcohol-free COD in both the chilled water receiver and the dry ice trap. The contents of the dry ice trap, however, contained three to four times the concentration of these components in the chilled water receiver. Organoleptic evaluation of these fractions is in progress.

SUMMARY OF COMPLETED STUDY OF AIR POLLUTION CREATED BY CITRUS FEED MILLS

John W. Seabury, P. E.

Executive Vice President
Fiske-Gay Associates, Inc.
Orlando, Florida

Under the sponsorship of the United States Department of Agriculture and with the cooperation of the Florida Cannery Association, a study has been made of air pollution resulting from operation of citrus pulp drying mills.

The study program was conducted with regard to type of machinery and method of operation for minimum air pollution and maximum operating efficiency.

Emissions to the atmosphere are regulated by the Florida Department of Pollution Control according to established statutory limits which are in compliance with National Standards.

Allowable emissions are based upon a graduated scale according to weight of material input to process.

With assistance of the United States Department of Agriculture and the Waste Committee of the Florida Cannery Association, three plants were chosen:

1. Tropicana Products, Inc., Bradenton, Florida.
2. Winter Garden Citrus Products Cooperative, Winter Garden, Florida.
3. Myakka Processors, Inc., Arcadia, Florida.

The Tropicana unit is a dryer-waste heat evaporator, wherein vapors from a furnace-dryer (with partial vapor recycle) serve as the heat source for a shell and tube molasses evaporator, and will be referred to as Type 1. This unit was manufactured by the Gulf Machinery Company of Clearwater, Florida.

The Winter Garden unit is a dryer-waste heat evaporator with reheat, wherein vapors from a furnace-dryer (with partial vapor recycle) serve as a heat source for a shell and tube molasses evaporator with reheat furnace for vapors after dryer, and will be referred to as Type 2. This unit was manufactured by Dan B. Vincent, Inc. of Tampa, Florida.

The Myakka unit is a dryer-scrubber, wherein vapors from a furnace-dryer (with partial vapor recycle) are passed through a two-stage water scrubber, and will be referred to as Type 3. This unit was manufactured by the Citrus By-Products Co., Lakeland, Florida.

Primary object of the study was to investigate processing variables as they might affect the amount and character of particulate matter emission from the stack at three different conditions for each mill. This requires a total of nine tests.

A total crew of five to eight men is needed for testing. Mill operations must be maintained at a steady rate during test with minimum variation of flows, moisture content, temperatures, etc.

Cooperation of plant management was excellent and with the obvious priority of keeping mill operation in step with peel production from juice operations, considerable skill was exercised in scheduling test dates when fruit supply would match test rate.

Wherever possible, quantity measurements were taken of peel input to process and press cake to dryer by means of platform scales and stopwatch. The same procedure was applied to finished product and to pellets if produced separately.

The screw conveyor being a steady flow device lends itself quite well to measurement, provided that a drop gate can be installed in a location with enough head room to allow for container and scale.

Some difficulty was encountered in getting 100% dropout with fully loaded high speed screws, as the material has a tendency to ride off-center and cling to flighting.

Size classification of finished product was done with mechanical shaker unit and five screen opening sizes: .371", .263", .185", .131", and .046". A 500 gram mill-run sample was placed in shaker and after five minutes of action the contents of each pan weighed and immediately placed in double plastic bags for later determination of moisture content. Classification runs were repeated at 15 to 20-minute intervals throughout the entire four to six hour test procedure. Moisture analysis of each size was performed in the laboratory by distillation with toluene according to the Association of Official Agricultural Chemists' method.

The technician at this station also recorded wet and dry bulb ambient temperatures and made spot checks of net bag weights of plant bagging equipment.

Condensate flows were measured by means of calibrated containers and stop watch. Liquid samples were placed on ice for laboratory determination of BOD and COD values. Likewise peel and press cake samples were taken at 20 to 30-minute intervals, double bagged, and placed on ice for later moisture test.

Particulate testing was carried out on stack emissions in accordance with procedures prescribed by the Florida Department of Pollution Control. Essentially, this consists of an accurate measurement of total quantity, painstaking analysis of a carefully withdrawn representative sample, and mathematical determination of the total emission by proportion; with compensation for temperature, pressure, and humidity.

The following Table gives condensed results from tests:

MILL TYPE	INPUT #/HR	MOISTURE REMOVAL #/HR	PRODUCT #/HR	CONDENSATE		MOISTURE %		EMISSION #/HR		FUEL		
				BOD	GPM	PEEL	PRODUCT	OBSERVED	ALLOWED	ft ³ /TON	#/TON	ft ³ /1000# WATER EVAP.
I	127,897	96,540	31,357	502	160	77.5	7.53	17.27	40.0	4,250	1.68	689.9
I	67,363	50,973	16,390	-	-	77.6	6.71	25.84	40.0	5,020	1.99	808.7
I	117,248	86,097	31,151	1721	162	75.8	8.69	6.20	40.0	4,670	1.86	845.6
II	32,057	25,817	6,240	783	33	81.8	6.53	14.23	23.47	8,280	3.51	1001.3
II	23,200	18,010	5,190	1020	26	78.8	5.12	18.88	18.12	10,170	4.34	1466.2
II	18,954	14,804	4,150	738	27	79.4	5.82	9.51	15.51	12,511	5.32	1753.6
III	16,814	13,534	3,280	0	0	81.7	6.08	75.35	14.24	95 GAL	6.17	11/5 GAL
III	17,936	14,946	2,990	0	0	85.0	10.02	38.90	14.56	87.5 GAL	5.70	9.8 GAL
III	14,721	10,661	4,058	0	0	75.4	10.44	8.63	12.96	*	*	*
										-	-	-

* Oil meter not working.

A total crew of five to eight men is needed for testing. Mill operations must be maintained at a steady rate during test with minimum variation of flows, moisture content, temperatures, etc.

Cooperation of plant management was excellent and with the obvious priority of keeping mill operation in step with peel production from juice operations, considerable skill was exercised in scheduling test dates when fruit supply would match test rate.

Wherever possible, quantity measurements were taken of peel input to process and press cake to dryer by means of platform scales and stopwatch. The same procedure was applied to finished product and to pellets if produced separately.

The screw conveyor being a steady flow device lends itself quite well to measurement, provided that a drop gate can be installed in a location with enough head room to allow for container and scale.

Some difficulty was encountered in getting 100% dropout with fully loaded high speed screws, as the material has a tendency to ride off-center and cling to flighting.

Size classification of finished product was done with mechanical shaker unit and five screen opening sizes: .371", .261", .185", .131", and .046". A 500 gram mill-run sample was placed in shaker and after five minutes of action the contents of each pan weighed and immediately placed in double plastic bags for later determination of moisture content. Classification runs were repeated at 15 to 20-minute intervals throughout the entire four to six hour test procedure. Moisture analysis of each size was performed in the laboratory by distillation with toluene according to the Association of Official Agricultural Chemists' method.

The technician at this station also recorded wet and dry bulb ambient temperatures and made spot checks of net bag weights of plant bagging equipment.

Condensate flows were measured by means of calibrated containers and stop watch. Liquid samples were placed on ice for laboratory determination of BOD and COD values. Likewise peel and press cake samples were taken at 20 to 30-minute intervals, double bagged, and placed on ice for later moisture test.

Particulate testing was carried out on stack emissions in accordance with procedures prescribed by the Florida Department of Pollution Control. Essentially, this consists of an accurate measurement of total quantity, painstaking analysis of a carefully withdrawn representative sample, and mathematical determination of the total emission by proportion; with compensation for temperature, pressure, and humidity.

The following Table gives condensed results from tests:

MILL TYPE	INPUT #/HR	MOISTURE REMOVAL #/HR	PRODUCT #/HR	CONDENSATE		MOISTURE %		EMISSION #/HR		FUEL		
				BOD	GPM	PEEL	PRODUCT	OBSERVED	ALLOWED	ft ³ /TON	#/TON	ft ³ /1000# WATER EVAP.
I	127,897	96,540	31,357	502	160	77.5	7.53	17.27	40.0	4,250	1.68	689.9
I	67,363	50,973	16,390	-	-	77.6	6.71	25.84	40.0	5,020	1.99	808.7
I	117,248	86,097	31,151	1721	162	75.8	8.69	6.20	40.0	4,670	1.86	845.6
II	32,057	25,817	6,240	783	33	81.8	6.53	14.23	23.47	8,280	3.51	1001.3
II	23,200	18,010	5,190	1020	26	78.8	5.12	18.88	18.12	10,170	4.34	1466.2
II	18,954	14,804	4,150	738	27	79.4	5.82	9.51	15.51	12,511	5.32	1753.6
III	16,814	13,534	3,280	0	0	81.7	6.08	75.35	14.24	95 GAL	6.17	11/5 GAL
III	17,936	14,946	2,990	0	0	85.0	10.02	38.90	14.56	87.5 GAL	5.70	9.8 GAL
III	14,721	10,661	4,058	0	0	75.4	10.44	8.63	12.96	* -	*	*

* Oil meter not working.

First cost is not easily comparable; individual circumstances and size differences make direct comparisons useful only on a very generalized basis.

The relative comparison finds total first cost is highest for Type 1, medium for Type 2, and least for Type 3.

On a basis of 1,000 pounds of water removed per hour first cost may vary from a low of \$10,000 up to \$17,000 depending on type of mill and amount of accessory machinery included.

IMPROVED METHOD FOR RECOVERY AND PURIFICATION OF ORANGE PEEL COLOR

C. W. Wilson, III, O. W. Bissett and R. E. Berry

Southeastern Marketing and Nutrition Research Division
Citrus and Subtropical Products Laboratory
Winter Haven, Florida

An improved method was developed for purification of color obtained by hexane extraction of orange peel. The method was tested on a pilot plant scale using peel from early and late maturity Hamlin, Temple, Pineapple and Valencia oranges and tangerines. The extraction was similar to that reported in previous citrus conference reports which included studies on the pilot scale hexane extraction of three types of commercially available citrus peel waste, (kilogram batches of flavedo, "frits," and ground whole peel). The hexane-color extracts were concentrated at reduced pressure. In the previous method crude color concentrates were vacuum steam-distilled to remove most of the volatile peel oil components, saponified with methanolic potassium hydroxide, and then neutralized with potassium acid phosphate. The color concentrates were then washed with a dilute water-ethanol solution to remove remaining traces of high boiling peel oil components. In those studies, yields up to 934, 630, and 538 mg color/kg peel were obtained for flavedo, frits, and ground whole peel, respectively. Additional color has also been obtained by acetone treatment of emulsions that formed during extraction and the total yield of color was thereby increased by 30% for flavedo, 50% for "frits," and 1% for ground whole peel.

In the current method the need for saponification, neutralization, and solvent-water washing was eliminated through a solvent precipitation procedure. After hexane extraction and vacuum steam distillation as previously described, the crude color concentrates were dissolved in 2-propanol and then diluted with water to give a final concentration of 60% 2-propanol and 40% water. This precipitated the carotenoids but left flavonoids and other impurities in solution. The precipitate was centrifuged or filtered from solution and dried under vacuum. The improved purification method shortened the total process time on a pilot scale by about 25%.

The improved process was applied to 180-lb units of FMC extractor whole peel and "frits" (ground peel and flavedo pieces expelled from an FMC In-line extractor), and to flavedo obtained from a Brown extractor and peel shaver. Color concentrates were prepared from two 180-lb units of each of the starting materials. Each 180-lb unit was prepared by extracting three 60 lb batches and combining the color extracts for subsequent processing. Yields of color were determined for each 180-lb. unit and averages were determined from duplicate determinations. Tangerine "frits" yielded 484 mg color/kg peel. Yields for early maturity Hamlins was 294 mg color/kg peel and for late maturity Hamlins 410 mg color/kg peel. Temple "frits" yielded 812 mg color/kg peel. Yields up to 1009, 366, and 424 mg color/kg peel were obtained from Pineapple flavedo, ground whole peel and "frits," respectively. Valencia flavedo, ground whole peel, and "frits" yielded 770, 593, and 367 mg color/kg peel, respectively. Material balance studies accounted for about 90% or more of the solids and indicated 80% hexane recovery.

The color enhancing capacity of these color concentrates was checked by addition to 45° Brix substandard early season Hamlin FCOJ, and by color reflectance instrumental determinations. In dilution studies all samples, regardless of source, effectively raised the USDA color score by one grade point when used at 1/6000. Results of instrumental analyses showed that tangerine "frits" were highest with respect to the three tristimulus values (i.e., degrees of redness, blueness and greenness). All other color concentrates, with the exception of those from Valencia ground whole peel and "frits," were about the same in tristimulus values. These values for color from Valencia ground whole peel and "frits" were somewhat lower. Flavor studies on orange juice concentrates with these color fractions added indicated the products were acceptable when the color was blended with essence and distilled oil or cold-pressed oil to make a highly colored oil and this was blended with the juice. Exceptions were the samples with color from Temples, tangerines and Hamlins, but these were satisfactory after such color fractions had been reprecipitated.

UNITED STATES DEPARTMENT OF AGRICULTURE

AGRICULTURAL RESEARCH SERVICE

LIST OF CITRUS PUBLICATIONS

AND PATENTS

(September 1, 1970 - August 31, 1971)

Reprints of publications may be obtained without cost by addressing request to the Laboratory listed.

Patents may be obtained only by purchase from the U. S. Patent Office, Washington, D. C. 20250, for 50 cents each.

SOUTHEASTERN MARKETING AND NUTRITION RESEARCH DIVISION

CITRUS AND SUBTROPICAL PRODUCTS LABORATORY

600 Avenue S, N. W.

Winter Haven, Florida 33880

GAS-LIQUID CHROMATOGRAPHY IN THE STUDY OF THE MAILLARD BROWNING REACTION

M. L. Wolf from and N. Kashimura

CONTRACTOR: The Ohio State University, Columbus, Ohio
Carbohyd. Res. 11, 151-152 (1969).

CITRUS PRODUCT COLOR ENHANCEMENT USING EXTRACTS OF PEEL OF DIFFERENT VARIETIES

T. J. Kew and R. E. Berry

J. Food Sci. 35: 436-439, 1970.

DEGRADATION OF D-GLUCOSE WITH ACETIC ACID AND METHYLAMINE

G. R. Jurch, Jr., and J. H. Tatum

Carbohyd. Res. 15: 233-239, 1970.

LIMONENE REDUCTASE SYSTEM IN THE ORANGE

V. H. Potty and J. H. Bruemmer

Phytochem. 9: 2319-2321, 1970.

NOOTKATENE FROM CITRUS SINENSIS

E. D. Lund, R. L. Coleman, and M. G. Moshonas

Phytochem. 9: 2419-2422, 1970.

LIMONIN IN FLORIDA CITRUS FRUITS

W. C. Scott

Proc. Fla. State Hort. Soc. 83: 270-277, 1970.

EFFECT OF TEMPERATURE ON CENTRIFUGAL SEPARATION OF COLD-PRESSED ORANGE OIL

R. E. Berry and P. Casals

Proc. Fla. State Hort. Soc. 83: 310-313, 1970.

BIOCHEMICAL CHANGES IN GRAPEFRUIT DURING ANAEROBIC METABOLISM

J. H. Bruemmer and B. Roe

Proc. Fla. State Hort. Soc. 83: 290-294, 1970.

ANALYSIS OF VOLATILE FLAVOR CONSTITUENTS FROM GRAPEFRUIT ESSENCE

M. G. Moshonas and P. E. Shaw

J. Agr. Food Chem. 19(1) 119-120, 1971.

2,3-DIHYDRO-3,5-DIHYDROXY-6-METHYL-4H-PYRAN-4-ONE, A DEGRADATION PRODUCT OF A HEXOSE

P. E. Shaw, J. H. Tatum and R. E. Berry

Carbohyd. Res. 16: 207-211, 1971.

TRAPPING VOLATILES FROM GLC FOR INJECTION INTO A MASS SPECTROMETER

M. G. Moshonas and P. E. Shaw

Appl. Spectry. 25(1) 101-102, 1971.

SUBSTRATE SPECIFICITY OF CITRUS ALCOHOL: NAD OXIDOREDUCTASE

J. H. Bruemmer and B. Roe

J. Agr. Food Chem. 19(2) 266-268, 1971.

ORANGE PEEL COLOR EXTRACT: ITS USE AND STABILITY IN CITRUS PRODUCTS

R. E. Berry, O. W. Bissett and T. J. Kew

J. Food Sci. 36: 367-369, 1971.

A GAS CHROMATOGRAPHIC PROCEDURE FOR ANALYSIS OF AQUEOUS ORANGE ESSENCE

M. G. Moshonas and E. D. Lund

J. Food Sci. 36(1) 105-106, 1971.

A RAPID GAS CHROMATOGRAPHIC METHOD FOR STUDYING VOLATILE CARBONYL COMPOUNDS FROM ORANGE JUICE AND THEIR CHANGES DURING STORAGE

H. L. Dinsmore and S. Nagy

J. Agr. Food Chem. 19(3) 517-519, 1971.

FATTY ACID PROFILES OF ORANGE AND TANGOR JUICE SAC LIPIDS

H. E. Nordby and S. Nagy

Phytochem. 10: 615-619, 1971.

PROCEEDINGS OF THE 1970 CONFERENCE ON CITRUS CHEMISTRY AND UTILIZATION

U. S. Dept. Agr., Agr. es. Serv., ARS 72-89: 33 pp., May 1971.

VITAMIN C RETENTION IN ORANGE JUICE AS RELATED TO CONTAINER TYPE

R. E. Berry, O. W. Bissett and M. K. Veldhuis

The Citrus Ind. 52(6) 12-13, 1971

ANALYSIS OF CARBONYL FLAVOR CONSTITUENTS FROM GRAPEFRUIT OIL

M. G. Moshonas

J. Agr. Food Chem. 19(4) 769-770, 1971.

ANALYSIS OF VALENCIA ORANGE ESSENCE AND AROMA OILS

R. L. Coleman and P. E. Shaw

J. Agr. Food Chem. 19(3) 520-523, 1971.

REGULATION OF NAD AND NADP OXIDOREDUCTASES IN ORANGES

J. H. Bruemmer and B. Roe

Phytochem. 10, 255-259, 1971.

DEVICE AND METHOD OF DIRECT INTRODUCTION OF MATERIALS INTO MASS SPECTROMETERS FROM ADSORBANTS

U. S. Patent No. 3,583,234, Patented June 8, 1971.

WESTERN MARKETING AND NUTRITION RESEARCH DIVISION

WESTERN REGIONAL RESEARCH LABORATORY

Berkeley, California 94710

LEMON-PEEL PECTIN. III. FRACTIONATION OF PECTIC ACIDS FROM LEMON-PEEL AND LUCERNE.

G. O. Aspinall, I. W. Cottrell, J. A. Molloy and M. Uddin.
Canadian Journal of Chemistry 48(8): 1290-95, April 15, 1970.
(P. L. 480 grant, University of Edinburgh, Scotland)

DER EFFEKT VON HITZE UND ⁶⁰CO-BESTRAHLUNG AUF DIE ASCOSPOREN VON BYSSOCHLAMYS FULVA IN VERSCHIEDENEN FRUCHTSAFTEN (The Effects of Heat and ⁶⁰Co-irradiation on the Ascospores of Byssochlamys fulva in Various Fruit Juices).

G. Partsch, O. Richter, and H. Altmann.
Mitt. Rebe und Wein Obstan und Fruchteverwertung 20: 234-40, 1970.
(P. L. 480 grant, Institute of Biology and Agriculture, Lenaugasse 10, Austria).

STUDIES ON CARBOHYDRATE METABOLISM IN FRUIT TISSUES OF CITRUS ACIDA
Part I. Detection of the Enzymes of the Glycolytic Cycle in the Fruit Tissues of Citrus Acida

Part II. Studies on Glycolytic Intermediates in the Fruit Tissues of Citrus acida During the Development of the Fruit

Part III. Ascorbic Acid Metabolism in Fruit Tissues of Citrus acida at Different Stages of the Development of the Fruit.

L. J. Parekh, K. K. Sakariah, and V. J. Shah.
Enzymologia 38(1): 19-32, 1970.
(P. L. 480 grant, M.S. University, Baroda, India).

RADIO RESISTANCE OF BYSSOCHLAMYS FULVA ASCOSPORES AS SHOWN BY STORAGE TESTS

G. Partsch and H. Altmann.
J. Food Sci. 35(3): 251-52, 1970
(P. L. 480 grant, Institute of Biology, Reactor Centre, Seibersdorf, Austria).

DIHYDROCHALCONE SWEETENERS

J. Peter Clark.
Proc. Soc. of Soft Drink Technologists, 17th, Orlando, Florida, May 4-6, 1970, pp. 101-104, Washington, D. C., 1970.

EFFECT OF TEMPERATURE ON STABILITY OF ORANGE AROMA SOLUTION

D. G. Guadagni, J. L. Bomben, and H. C. Mannheim.
J. Food Sci. 35(3): 279-81, May-June 1970.

STORAGE STABILITY OF FROZEN ORANGE JUICE CONCENTRATE MADE WITH AROMA SOLUTION OR CUTBACK JUICE.

D. G. Guadagni, J. L. Bomben, and H. C. Mannheim.
Food Technol. 24(9): 1012-16, Sept. 1970.

ODOUR CHARACTER AND THRESHOLD VALUES OF NOOTKATONE AND RELATED COMPOUNDS.

K. L. Stevens, D. G. Guadagni, and D. J. Stern

J. Sci. Food Agr. 21: 590-93, Nov. 1970.

PHOTOLYSIS OF NOOTKATONE--AN UNUSUAL INTRAMOLECULAR PHOTOCYCLISATION.

K. L. Stevens.

J. Sci. Food Agr. 21: 593-96, Nov. 1970.

LIQUID CARBON DIOXIDE FOR SELECTIVE AROMA EXTRACTION.

W. G. Schultz and J. M. Randall

Food Technol. 24(11): 94-98, 1970.

DYNAMIC TURBULENCE PROMOTION IN REVERSE OSMOSIS PROCESSING OF LIQUID FOODS

E. Lowe and E. L. Durkee.

J. Food Sci. 36: 31-32, 1971.

SOLID-LIQUID PHASE EQUILIBRIA IN MULTICOMPONENT AQUEOUS SUGAR SOLUTIONS

S. K. Chandrasekaran and C. Judson King.

J. Food Sci. 36: 699-704, 1971.

(Research done under grant to University of California, Berkeley).

THE ANTIMICROBIAL ACTIVITY OF CITRAL

K. L. Stevens, L. Jurd, A. D. King, Jr., and K. Mihara.

Experimentia 27: 600-2, 1971.

CITRIC ACID METABOLISM IN THE FRUIT TISSUES OF CITRUS ACIDA

C. V. Ramakrishnan.

Curr. Sci. 40(5): 97-100, March 5, 1971.

(P. L. 480 grant, M. S. University, Baroda, India).

WESTERN MARKETING AND NUTRITION RESEARCH DIVISION

FRUIT AND VEGETABLE CHEMISTRY LABORATORY

263 South Chester Avenue
Pasadena, California 91106

CITRUS JUICE CHARACTERIZATION. II. IDENTIFICATION AND ESTIMATION OF
THE MAJOR PHOSPHOLIPIDS

Carl E. Vandercook, Henry C. Guerrero, and Ruth L. Price
J. Agr. Food Chem. 18(5): 905-7, 1970

CHANGES IN ULTRAVIOLET SPECTRAL PROPERTIES OF LEMON JUICE UNDER ADVERSE
STORAGE CONDITIONS

Carl E. Vandercook
J. Food Sci. 35: 517-18, 1970

BIOSYNTHESIS OF TRANS-CINNAMATE FROM PHENYLPYRUVATE AND L-GLUTAMATE BY
CELL-FREE EXTRACTS OF GRAPEFRUIT

Shin Hasegawa and V. P. Maier
Phytochem. 9: 2483-87, 1970.

CAROTENONE FORMATION IN TRIPHASIA TRIFOLIA

Henry Yokoyama and Michael J. White
Phytochem. 9(8): 1795-97, 1970.

PHENYLALANINE AMMONIA-LYASE ACTIVITY IN CITRUS FRUIT TISSUE CULTURED
IN VITRO

T. A. Thorpe, V. P. Maier, and Shin Hasegawa
Phytochem. 10: 711-18, 1971.

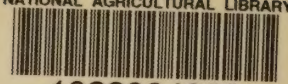
DEVELOPMENT OF METHODS FOR PRODUCING NONBITTER NAVEL ORANGE JUICE

V. P. Maier, L. C. Brewster, and A. C. Hsu
Citrograph 56: 351, 1971.

ENZYME PREPARATION OF HESPERETIN DIHYDROCHALCONE GLUCOSIDE

R. M. Horowitz and B. Gentili
U. S. Patent 3,583,894. June 8, 1971.

NATIONAL AGRICULTURAL LIBRARY



1022934968